Claims

What is claimed is:

1. A natriuretic compound conjugate comprising:

(a) a biologically active natriuretic compound comprising:

(i) a natriuretic molecule NPR-A binding site; and

(ii) at least one modifying moiety conjugation site; and

(b) at least one modifying moiety attached to said modifying moiety conjugation site;

wherein said natriuretic compound conjugate exhibits one or more advantages selected from the group consisting of increased resistance to enzymatic degradation relative to a corresponding unconjugated natriuretic compound, increased circulating half life, increased bioavailability, and prolonged duration of effect.

2. The natriuretic compound conjugate of claim 1 further defined as retaining a therapeutically significant percentage of cGMP stimulating activity relative to the corresponding unconjugated natriuretic compound.

3. The natriuretic compound conjugate of claim 1 further defined as retaining at least 30% of the cGMP stimulating activity of the corresponding unconjugated natriuretic compound.

4. The natriuretic compound conjugate of claim 1 further defined as retaining at least 50% of the cGMP stimulating activity of the corresponding unconjugated natriuretic compound.

5. The natriuretic compound conjugate of claim 1 further defined as retaining at least 70% of the cGMP stimulating activity of the corresponding unconjugated natriuretic compound.

6. The natriuretic compound conjugate of claim 1 further defined as retaining at least 90% of the cGMP stimulating activity of the corresponding unconjugated natriuretic compound.

7. The natriuretic compound conjugate of claim 1 further defined as more hydrophilic than a corresponding unconjugated natriuretic compound.

8. The natriuretic compound conjugate of claim 1 further defined as more amphiphilic than a corresponding unconjugated natriuretic compound.

9. The natriuretic compound conjugate of claim 1 further defined as more lipophilic than a corresponding unconjugated natriuretic compound.

10. The natriuretic compound conjugate of claim 9 wherein the modifying moiety does not consist of an alkyl moiety.

11. The natriuretic compound conjugate of claim 1 further defined as more resistant to protease degradation than a corresponding unconjugated natriuretic compound.

12. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises a sequence:

$A^1PX^1MVQGSGCFGRX^2MDRISSSSGLGCX^3VLR$ (SEQ ID NO. __).

wherein

A^1 is an amino acid or series of amino acids native to a natriuretic peptide,

X^1, X^2 and X^3 are independently selected from the group consisting of Lys, Arg and Gly, and at least one of X^1, X^2 and X^3 is a Lys.

13. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises a peptide or a biologically active peptide segment of brain natriuretic peptide, atrial natriuretic peptide, C-type natriuretic peptide, or dendroaspis natriuretic peptide.

14. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises:

(a) an amino acid sequence

$X^1 - C^1FGRX^2MDRISSSSGLGC^2 - X^3$ (SEQ ID NO:)

wherein

X^1 is optionally present and when present is an amino acid sequence having from 1-10 amino acids;

X^2 is Gly, Arg, or Lys; and

X^3 is optionally present and when present is an amino acid sequence having from 1-10 amino acids.

(b) a disulfide bond between C^1 and C^2 to form a loop.

15. The natriuretic compound conjugate of claim 14 wherein X^1 is Arg or Gly.

16. The natriuretic compound conjugate of claim 14 wherein X^1 is selected from the group consisting of:

(a) Lys;

(b) Gly;

(c) Arg;

(d) SG- (SEQ ID NO. __), GSG- (SEQ ID NO. __), QGSG- (SEQ ID NO. __), VQGSG- (SEQ ID NO. __), MVQGSG- (SEQ ID NO. __), PKMVQGSG- (SEQ ID NO. __), and SPKMVQGSG- (SEQ ID NO. __);

(e) hBNP segments of (d) comprising a substitution selected from the group consisting of Lys-to-Gly and Lys-to-Arg;

(f) hBNP segments of (d) comprising a substitution selected from the group consisting of Gly-to-Lys and Arg-to-Lys;

(g) hBNP segments of (d) comprising an inserted Lys;

(h) N-terminal tails and C-terminal segments of N-terminal tails of natriuretic peptides;

(i) N-terminal tails and C-terminal segments of (h) comprising a substitution selected from the group consisting of Lys-to-Gly and Lys-to-Arg;

(j) N-terminal tails and C-terminal segments of (h) comprising a substitution selected from the group consisting of Gly-to-Lys and Arg-to-Lys;

(k) N-terminal tails and C-terminal segments of (h) comprising an inserted Lys.

17. The natriuretic compound conjugate of claim 14 wherein X^3 is selected from the group consisting of:

(a) Lys;

(b) Gly;

(c) Arg;

(d) hBNP segments KV(SEQ ID NO. __), KVL (SEQ ID NO. __), KVLR (SEQ ID NO. __), KVLRR (SEQ ID NO. __), and KVLRRH (SEQ ID NO. __); and

(e) hBNP segments of (d) comprising a substitution selected from the group consisting of Lys-to-Gly and Lys-to-Arg;

(f) hBNP segments of (d) comprising a substitution selected from the group consisting of Gly-to-Lys and Arg-to-Lys;

(g) hBNP segments of (d) comprising an inserted Lys;

(h) C-terminal tails and N-terminal segments of C-terminal tails of natriuretic peptides;

(i) C-terminal tails and N-terminal segments of C-terminal tails of (h) comprising a substitution selected from the group consisting of Lys-to-Gly and Lys-to-Arg;

(j) C-terminal tails and N-terminal segments of C-terminal tails of (h) comprising a substitution selected from the group consisting of Gly-to-Lys and Arg-to-Lys;

(k) C-terminal tails and N-terminal segments of C-terminal tails of (h) comprising an inserted Lys.

18. The natriuretic compound conjugate of claim 14 wherein the natriuretic compound comprises a sequence selected from the group consisting of:

(a) SPKMVQGSGCFGRKMDRISSSSGLGCKVL (SEQ ID NO. __);

(b) SPKMVQGSGCFGRKMDRISSSSGLGC (SEQ ID NO. __); and

(c) segments (a) or (b) comprising a substitution selected from the group consisting of Lys-to-Gly and Lys-to-Arg .

19. The natriuretic compound conjugate of claim 14 wherein X^1 comprises a 1-9 amino acid residue sequence from the N-terminus of hBNP.

20. The natriuretic compound conjugate of claim 14 wherein X^1 comprises $SPX^3MVQGSG$ (SEQ ID NO:), and wherein X^2 comprises a modifying moiety conjugation site.

21. The natriuretic compound conjugate of claim 14 wherein X^3 comprises a 1-6 amino acid residue sequence from the C-terminus of hBNP.

22. The natriuretic compound conjugate of claim 14 wherein X^3 comprises KVLRRH (SEQ. ID. NO:), KVLRR ( ), KVLR ( ), KVL, KV or K.

23. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises a native hBNP sequence (SEQ ID NO. __) having one or more mutations selected from the group consisting of Lys3Arg, Lys14Arg, Arg30Lys, Lys27Arg, and Arg31Lys.

24. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises a native hBNP sequence (SEQ ID NO. __), having one or more insertions or deletions.

25. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises a native hBNP amino acid sequence (SEQ ID NO. __) and a N-terminal or C-terminal Lys.

26. The natriuretic compound conjugate of claim 1 further defined as:

(a) comprising a multipeptide comprising two or more amino acid sequences encoding a natriuretic compound;

(b) optionally comprising a spacer sequence between each set or adjacent natriuretic compound encoding sequences;

(c) optionally comprising an extension at either or both ends of the multipeptide, the extension comprising one or more amino acids.

27. The natriuretic compound conjugate of claim 26 wherein the natriuretic peptide units each comprise hBNP (SEQ ID NO. __) or a biologically active analog, segment or segment analog thereof.

28. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound consists of a native BNP.

29. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound consists of a native hBNP.

30. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound consists of a native ANP.

31. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound consists of a canine BNP.

32. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound consists of urodilatin.

33. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound consists of DNP.

34. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises an amino acid sequence:

X^1MVQGSGCFGRX^2MDRISSSSGLGCX^3 (SEQ ID NO. __),

wherein X^1, X^2 and X^3 are each independently selected from the group consisting of Lys, Gly and Arg, with the proviso that at least one of X^1, X^2 and X^3 is Arg or Gly.

35. The natriuretic compound conjugate of claim 34 wherein the sequence comprises:

(a) N-terminal to X^1, an extension selected from the group consisting of: SPK, PK and K; and/or

(b) C-terminal to X^3, an extension selected from the group consisting of -VLRRH (SEQ ID NO:), -VLRR (SEQ ID NO:), -VLR, -VL, and -V.

36. The natriuretic compound conjugate of claim 34 wherein X^1 is Lys, X^2 is Arg and X^3 is Arg.

37. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises an amino acid sequence:

$CFGRX^1MDRISSSSGLGCX^2$ (SEQ ID NO:),

wherein X^1 and/or X^2 comprises a modifying moiety conjugation site coupled to the modifying moiety.

38. The natriuretic compound conjugate of claim 37 wherein X^1 comprises Lys coupled to the modifying moiety.

39. The natriuretic compound conjugate of claim 37 wherein X^2 comprises Lys coupled to the modifying moiety.

40. The natriuretic compound conjugate of claim 1 wherein the modifying moiety conjugation site comprises a moiety selected from the group consisting of natural or non-natural amino acid side chains, an N-terminus of the natriuretic compound, and a C-terminus of the natriuretic compound.

41. The natriuretic compound conjugate of claim 40 wherein the modifying moiety conjugation site is a Lys side chain.

42. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound conjugate includes only one modifying moiety.

43. The natriuretic compound conjugate of claim 1 wherein:

(a) the natriuretic compound comprises a Lys^3 to Cys^{26} segment of hBNP and a disulfide bond coupling Cys^{10} of the segment to the Cys^{26} (SEQ ID NO. __);

(b) a single modifying moiety coupled to the natriuretic compound at the Lys^3 (SEQ ID NO. __).

44. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises a Cys^{10} to Cys^{26} segment of hBNP and a disulfide bond coupling the Cys^{10} to the Cys^{26}, wherein said natriuretic compound is a monoconjugate including a single modifying moiety coupled thereto at Lys^{14} of the segment.

45. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises a Cys^{10} to Lys^{27} segment of hBNP (SEQ ID NO. __), wherein said natriuretic compound is a monoconjugate including a single modifying moiety coupled thereto at Lys^{27} of the segment.

46. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises a Cys^{10} to His^{32} segment of hBNP and a disulfide bond coupling the Cys^{10} to Cys^{26} of the segment, wherein said natriuretic compound is a monoconjugate including a single modifying moiety coupled thereto at Lys^{27} of the segment.

47. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound comprises a Cys^{10} to Cys^{26} segment of hBNP and a disulfide bond coupling the Cys^{10} to the Cys^{26}; wherein the natriuretic compound is a monoconjugate including a single modifying moiety coupled thereto at the N-terminus of the natriuretic compound.

48. The natriuretic compound conjugate of claim 1 wherein:

(a) the natriuretic compound consists of the hBNP amino acid sequence; and

(b) the natriuretic compound conjugate is a diconjugate comprising:

(i) a modifying moiety coupled to the natriuretic peptide at Lys^3 of the hBNP amino acid sequence; and

(ii) a modifying moiety coupled to the natriuretic peptide at Lys^{14} of the hBNP amino acid sequence.

49. The natriuretic compound conjugate of claim 1 wherein:

(a) the natriuretic compound is hBNP; and

(b) the natriuretic compound conjugate is a diconjugate comprising:

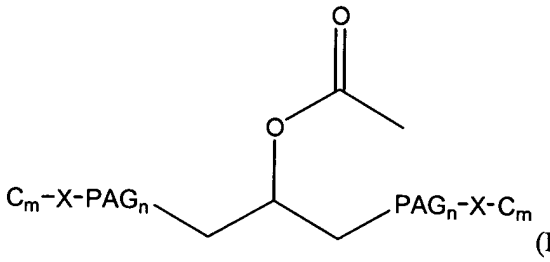
(i) a modifying moiety coupled to the natriuretic peptide at Lys^3 of the hBNP amino acid sequence; and

(ii) a modifying moiety coupled to the natriuretic peptide at Lys^{27} of the hBNP amino acid sequence.

50. The natriuretic compound conjugate of claim 1 wherein the natriuretic compound sequence comprises an N-terminal tail and the modifying moiety is coupled to an amino acid which is positioned in the N-terminal tail.

51. The natriuretic compound conjugate of claim 50 wherein the N-terminal tail consists of a native sequence of an N-terminal tail of a natriuretic peptide or a C-terminal segment of an N-terminal tail of a natriuretic peptide.

52. The natriuretic compound conjugate of claim 1 wherein the modifying moiety has a formula:



(Formula I)

wherein

each C is independently selected and is an alkyl moiety having m carbons and m is from 1 to 20; and

each PAG is independently selected and is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

each X is independently selected and is a linking moiety.

53. The natriuretic compound conjugate of claim 52 wherein m is from 1 to 18.

54. The natriuretic compound conjugate of claim 52 wherein m is from 1 to 16.

55. The natriuretic compound conjugate of claim 52 wherein n is from 2 to 20.

56. The natriuretic compound conjugate of claim 52 wherein n is from 2 to 15.

57. The natriuretic compound conjugate of claim 52 wherein n is from 2 to 10.

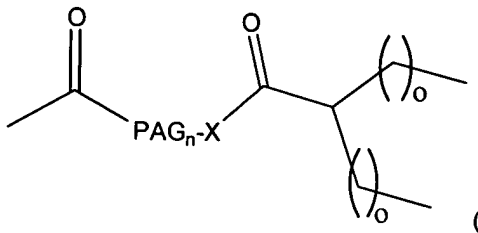
58. The natriuretic compound conjugate of claim 52 wherein each X is independently selected from the group consisting of -C-, -O-, -C(O)-, -NH-, -NHC(O)-, and -C(O)NH-.

59. The natriuretic compound conjugate of claim 52 wherein the modifying moiety renders the natriuretic compound more lipophilic than a corresponding unconjugated natriuretic compound.

60. The natriuretic compound conjugate of claim 52 wherein the modifying moiety renders the natriuretic compound more hydrophilic than a corresponding unconjugated natriuretic compound.

61. The natriuretic compound conjugate of claim 52 wherein the modifying moiety renders the natriuretic compound more amphiphilic than a corresponding unconjugated natriuretic compound.

62. The natriuretic compound conjugate of claim 1 wherein the modifying moiety has a formula:



(Formula II)

PAG is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

X is O or N; and

each o is independently selected and is from 1 to 15.

63. The natriuretic compound conjugate of claim 62 wherein n is from 2 to 20.

64. The natriuretic compound conjugate of claim 62 wherein n is from 2 to 15.

65. The natriuretic compound conjugate of claim 62 wherein n is from 2 to 10.

66. The natriuretic compound conjugate of claim 62 wherein each o is independently selected and is from 1 to 13.

67. The natriuretic compound conjugate of claim 62 wherein each o is independently selected and is from 1 to 9.

68. The natriuretic compound conjugate of claim 62 wherein each o is independently selected and is from 1 to 6.

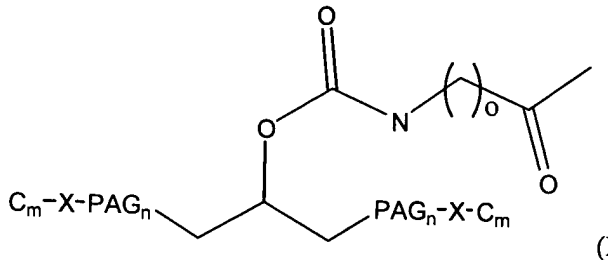
69. The natriuretic compound conjugate of claim 62 wherein each X is -O-.

70. The natriuretic compound conjugate of claim 62 wherein the modifying moiety renders the natriuretic compound more lipophilic than a corresponding unconjugated natriuretic compound.

71. The natriuretic compound conjugate of claim 62 wherein the modifying moiety renders the natriuretic compound more hydrophilic than a corresponding unconjugated natriuretic compound.

72. The natriuretic compound conjugate of claim 62 wherein the modifying moiety renders the natriuretic compound more amphiphilic than a corresponding unconjugated natriuretic compound.

73. The natriuretic compound conjugate of claim 1 wherein the modifying moiety has a formula:



(Formula III)

each C is independently selected and is an alkyl moiety having m carbons and m is from 1 to 20; and

each PAG is independently selected and is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

each X is independently selected and is a linking moiety.

o is from 1 to 15.

74. The natriuretic compound conjugate of claim 73 wherein m is from 1 to 18.

75. The natriuretic compound conjugate of claim 73 wherein m is from 1 to 16.

76. The natriuretic compound conjugate of claim 73 wherein n is from 2 to 20.

77. The natriuretic compound conjugate of claim 73 wherein n is from 2 to 15.

78. The natriuretic compound conjugate of claim 73 wherein n is from 2 to 10.

79. The natriuretic compound conjugate of claim 73 wherein o is from 1 to 13.

80. The natriuretic compound conjugate of claim 73 wherein o is from 1 to 9.

81. The natriuretic compound conjugate of claim 73 wherein o is from 1 to 6.

82. The natriuretic compound conjugate of claim 73 wherein each X is independently selected from the group consisting of -C-, -O-, -C(O)-, -NH-, -NHC(O)-, and -C(O)NH-.

83. The natriuretic compound conjugate of claim 73 wherein the modifying moiety renders the natriuretic compound more lipophilic than a corresponding unconjugated natriuretic compound.

84. The natriuretic compound conjugate of claim 73 wherein the modifying moiety renders the natriuretic compound more hydrophilic than a corresponding unconjugated natriuretic compound.

85. The natriuretic compound conjugate of claim 73 wherein the modifying moiety renders the natriuretic compound more amphiphilic than a corresponding unconjugated natriuretic compound.

86. The natriuretic compound conjugate of claim 1 wherein the modifying moiety comprises a linear or branched polyalkylene glycol moiety.

87. The natriuretic compound conjugate of claim 1 wherein the modifying moiety comprises a sugar moiety coupled to an alkyl moiety.

88. The natriuretic conjugate of claim 87 wherein the modifying moiety further comprises a sugar moiety.

89. The natriuretic compound conjugate of claim 86 wherein the polyalkylene glycol moiety comprises a polyethylene glycol moiety.

90. The natriuretic compound conjugate of claim 86 wherein the polyalkylene glycol moiety has from 2 to 25 polyalkylene glycol subunits.

91. The natriuretic compound conjugate of claim 86 wherein the polyalkylene glycol moiety has from 2 to 20 polyalkylene glycol subunits.

92. The natriuretic compound conjugate of claim 86 wherein the polyalkylene glycol moiety has from 2 to 15 polyalkylene glycol subunits.

93. The natriuretic compound conjugate of claim 86 wherein the polyalkylene glycol moiety has from 2 to 10 polyalkylene glycol subunits.

94. The natriuretic compound conjugate of claim 86 wherein the modifying moiety further comprises a linear or branched alkyl moiety.

95. The natriuretic compound conjugate of claim 94 wherein the modifying moiety further comprises a sugar moiety.

96. The natriuretic compound conjugate of claim 94 wherein the alkyl moiety has from 1 to 20 carbons.

97. The natriuretic compound conjugate of claim 94 wherein the alkyl moiety has from 1 to 18 carbons.

98. The natriuretic compound conjugate of claim 94 wherein the alkyl moiety has from 1 to 16 carbons.

99. The natriuretic compound conjugate of claim 94 wherein the alkyl moiety is separated from the polyalkylene glycol moiety by a linker selected from the group consisting of -C-, -O-, -C(O)-, -NH-, -NHC(O)-, and -C(O)NH-.

100. The natriuretic compound conjugate of claim 94 wherein the modifying moiety renders the natriuretic compound conjugate more lipophilic than a corresponding unconjugated natriuretic compound.

101. The natriuretic compound conjugate of claim 94 wherein the modifying moiety comprises a bond coupling the polyalkalene glycol moiety to the alkyl moiety which bond is hydrolysable *in vivo*.

102. The natriuretic compound conjugate of claim 1 wherein the modifying moiety comprises a linear or branched polyalkylene glycol moiety coupled to the natriuretic compound and a linear or branched alkyl moiety coupled to the polyalkalene glycol moiety at a site which is distal relative to the natriuretic compound.

103. The natriuretic compound conjugate of claim 1 wherein the modifying moiety comprises a linear or branched alkyl moiety coupled to the natriuretic compound and a polyalkylene glycol moiety coupled to the alkyl moiety at a site which is distal relative to the natriuretic compound.

104. The natriuretic compound conjugate of claim 1 wherein the modifying moiety is selected from the group consisting of the oligomeric moieties of **Table 1**.

105. The natriuretic compound conjugate of claim 1 wherein the modifying moiety is coupled to the natriuretic compound by a bond that is hydrolysable *in vivo*.

106. The natriuretic compound conjugate of claim 1 wherein the modifying moiety is coupled to the natriuretic compound by a bond that is hydrolysable in the bloodstream.

107. The natriuretic compound conjugate of claim 1 wherein the modifying moiety is coupled to the natriuretic compound by a bond that is not hydrolysable *in vivo*.

108. The natriuretic compound conjugate of claim 1 wherein the modifying moiety is coupled to the natriuretic compound by a bond that is not hydrolysable in the bloodstream.

109. The natriuretic compound conjugate of claim 1 wherein the modifying moiety is coupled to the natriuretic compound by a bond selected from the group consisting of ester, carbonate, carbamate, amide, ether, and amine.

110. The natriuretic compound conjugate of claim 1 wherein the modifying moiety is hydrolysable *in vivo* to yield a pegylated natriuretic compound.

111. The natriuretic compound conjugate of claim 110 wherein the modifying moiety is hydrolysable *in vivo* to yield a pegylated natriuretic compound comprising one or more PEG moieties having from 1 to 6 PEG units.

112. A pharmaceutical formulation comprising the natriuretic compound conjugate of claim 1.

113. The pharmaceutical formulation of claim 112 formulated for a route of delivery selected from the group consisting of enteral, perenteral, oral, subcutaneous, sublingual, buccal, nasal, intravenous and intramuscular.

114. A method of treating a condition characterized by an excessive level of extracellular fluid, the method comprising administering to a subject in need thereof a pharmaceutically acceptable amount of a natriuretic compound conjugate of claim 1.

115. The method of claim 114 wherein the condition comprises congestive heart failure.

116. The method of claim 114 wherein the condition comprises chronic congestive heart failure.

117. The method of claim 114 wherein the condition comprises acute congestive heart failure.

118. The method of claim 114 wherein the natriuretic compound conjugate is self-administered.

119. The method of claim 114 wherein the natriuretic compound conjugate is orally administered.

120. The method of claim 114 wherein the natriuretic compound conjugate is administered via a route of administration selected from the group consisting of enteral, perenteral, oral, subcutaneous, sublingual, buccal, nasal, intravenous and intramuscular.

121. The method of claim 114 wherein the condition is hypertension.

122. A method of making the natriuretic compound conjugate of claim 1, the method comprising:

(a) conjugating a natriuretic peptide multipeptide comprising two or more natriuretic compound units;

(b) cleaving the natriuretic peptide multipeptide to yield natriuretic compound conjugate;

(c) oxidizing the cleaved natriuretic compound conjugate to form one or more disulfide bonds in the natriuretic compound conjugate.

123. The method of claim 122 wherein the natriuretic compound comprises Cys^{10} to Cys^{26} of hBNP (SEQ ID NO. __) and step 122(c) yields a disulfide bond between the Cys^{10} and Cys^{26}.

124. A method of making the natriuretic compound conjugate of claim 1, the method comprising:

(a) making a multi-peptide natriuretic compound comprising two or more natriuretic compound units;

(b) cleaving the natriuretic peptide multipeptide to yield natriuretic peptide compound;

(c) conjugating the natriuretic compound to yield natriuretic compound conjugate;

(d) oxidizing the cleaved natriuretic compound conjugate to form one or more disulfide bonds in the natriuretic compound conjugate.

125. The method of claim 124 wherein the natriuretic compound comprises Cys^{10} to Cys^{26} of hBNP (SEQ ID NO. __) and step 122(c) yields a disulfide bond between the Cys^{10} and Cys^{26}.

126. A method of making the natriuretic compound conjugate of claim 1, the method comprising:

(a) making a multi-peptide natriuretic compound comprising two or more natriuretic compound units;

(b) cleaving the natriuretic peptide multipeptide to yield natriuretic compound;

(c) oxidizing the cleaved natriuretic compound to form one or more disulfide bonds in the natriuretic compound; and

(d) conjugating the natriuretic compound.

127. A modified pro-polynatriuretic peptide conjugate comprising:

(a) at least one natureteic peptide unit having a modifying moiety conjugation site and an NPR-A binding site;

(b) at least one modifying moeity attached to the modifying moiety conjugation site of at least one of the natriuretic peptide units;

(c) a leader sequence; and

(d) an enzymatically cleavable spacer coupling the leader sequence to a first natriuretic peptide conjugate.

128. A natriuretic peptide analog comprising an amino acid sequence having at least one modifying moiety conjugation site, an NPR-A binding region and at least one substituted Lys residue therein as compared to a native natriuretic peptide amino acid sequence, wherein said substituted Lys residue is not the amino acid modifying moiety conjugation site.

129. The natriuretic peptide analog of claim 128 wherein the one or more substituted Lys residues comprise a substitution selected from the group consisting of: Lys3Gly, Lys3Arg, Lys14Gly, Lys14Arg, Lys27Gly, or Lys27Arg.

130. The natriuretic peptide analog of claim 128 comprising a structure:

SPKMVQGSGCFGRX^{1}MDRISSSSGLGCX^{2}VLRRH (SEQ ID NO:)

wherein X^1 is Lys and X^2 is other than Lys, or X^1 is Lys and X^2 is other than Lys, or X^1 and X^2 are other than Lys.

131. The natriuretic peptide analog of claim 130 wherein X^1 is Lys and X^2 is Arg or Gly, or X^1 is Lys and X^2 is Arg or Gly, or X^1 and X^2 are independently selected and are Arg or Gly.

132. A natriuretic peptide analog comprising a structure:

CFGRX^{1}MDRISSSSGX^{2}GC (SEQ ID NO:)

wherein X^1 is an amino acid that does not comprise a conjugation site, and X^2 is an amino acid that comprises a modifying moiety conjugation site.

133. The natriuretic peptide analog of claim 132 wherein X^1 is Arg and X^2 is Lys.

134. A natriuretic peptide analog having a structure:

X^1-CFGRX^3MDRISSSSGLGC-X^2 (SEQ ID No.)

wherein X^1 is an amino acid sequence having from 1 to 10 amino acids, X^2 is an amino acid sequence having from 1 to 10 amino acids, and X^3 is other than Lys.

135. The natriuretic peptide analog of claim 134 wherein X^3 is Arg or Gly.

136. The natriuretic peptide analog of claim 134 wherein X^1 is SPY^1MVQGSG (SEQ ID NO:), wherein Y^1 comprises a modifying moiety conjugation site.

137. The natriuretic peptide analog of claim 134 wherein X^1 is selected from the group consisting of:

(a) N-terminal tails and C-terminal segments of N-terminal tails of natriuretic peptides;

(b) N-terminal tails and C-terminal segments of (a) comprising a substitution selected from the group consisting of Lys-to-Gly and Lys-to-Arg;

(c) N-terminal tails and C-terminal segments of (a) comprising a substitution selected from the group consisting of Gly-to-Lys and Arg-to-Lys;

(d) N-terminal tails and C-terminal segments of (a) comprising an inserted Lys.

138. The natriuretic peptide analog of claim 134 wherein X^2 is Y^2VLRRH (SEQ. ID. NO:), wherein Y^2 is other than Lys.

139. The natriuretic peptide analog of claim 138 wherein Y^2 is Arg.

140. The natriuretic peptide analog of claim 134 wherein X^2 is selected from the group consisting of:

(a) C-terminal tails and N-terminal segments of C-terminal tails of natriuretic peptides;

(b) C-terminal tails and N-terminal segments of C-terminal tails of 137(a) comprising a substitution selected from the group consisting of Lys-to-Gly and Lys-to-Arg;

(c) C-terminal tails and N-terminal segments of C-terminal tails of 137(a) comprising a substitution selected from the group consisting of Gly-to-Lys and Arg-to-Lys;

(d) C-terminal tails and N-terminal segments of C-terminal tails of 137(a) comprising an inserted Lys.

141. A natriuretic peptide analog having a structure:

X^1-CFGRX^3MDRIGLGC-X^2 (SEQ ID No.)

wherein X^1 is a peptide of from 1 to 9 amino acids, X^2 is a peptide of from 1 to 6 amino acids, and X^3 is other than Lys.

142. The natriuretic peptide analog of claim 140 wherein X^3 is Arg or Gly.

143. The natriuretic peptide analog of claim 142 wherein X^1 is SPY^1MVQGSG (SEQ ID NO:), wherein Y^1 comprises a modifying moiety conjugation site.

144. The natriuretic peptide analog of claim 142 wherein X^2 is Y^2VLRRH (SEQ. ID. NO:), wherein Y^2 is other than Lys.

145. The natriuretic peptide analog of claim 144 wherein Y^2 is Arg.

146. The natriuretic peptide analog of claim 144 wherein X^3 is Arg, X^1 is a sequence SPKMVQGSG (SEQ ID NO:) and X^2 is a sequence RVL.

147. A natriuretic peptide analog having a structure X^1-CFGRX^3MDRIX^4GLGC-X^2 wherein

(a) X^1 is an amino acid sequence of from 1 to 10 amino acids,

(b) X^2 is an amino acid sequence of from 1 to 10 amino acids,

(c) X^4 is an amino acid sequence of from 1 to 4 amino acids; and

(d) X^3 is other than Lys.

148. The natriuretic peptide analog of claim 147 wherein neither X^1 nor X^2 is a sequence native to a natriuretic peptide.

149. The natriuretic peptide of claim 147 where X^3 is Arg or Gly.

150. The natriuretic peptide of claim 147 where X^1 is $SPY^1MVQGSG$ (SEQ ID NO: ____) wherein Y^1 comprises a modifying moiety conjugation site.

151. The natriuretic peptide analog of claim 147 wherein X^2 is Y^2VLRRH (SEQ. ID. NO:), wherein Y^2 is other than Lys.

152. The natriuretic peptide analog of claim 151 wherein Y^2 is Arg.

153. An hBNP analog comprising a substitution of Lys14Arg or Lys14Gly.

154. An hBNP analog comprising a substitution of Lys27Arg or Lys27Gly.

155. An hBNP analog comprising a substitution of Lys3Arg or Lys3Gly.

156. A natriuretic compound conjugate comprising:

(a) a natriuretic compound comprising:

(i) a natriuretic molecule NPR-A binding site; and

(ii) at least one modifying moiety conjugation site; and

(b) at least one modifying moiety attached to said modifying moiety conjugation site;

wherein said natriuretic compound retains a therapeutically significant percentage of cGMP stimulating activity relative to a corresponding unconjugated natriuretic compound.

157. A natriuretic compound conjugate comprising:

(a) a natriuretic compound comprising:

(i) a natriuretic molecule NPR-A binding site; and

(ii) at least one modifying moiety conjugation site; and

(b) at least one modifying moiety attached to said modifying moiety conjugation site;

wherein said natriuretic compound conjugate retains at least 50% of the cGMP stimulating activity of a corresponding unconjugated natriuretic compound.

158. A natriuretic compound conjugate comprising:

(a) a natriuretic compound comprising:

(i) a natriuretic molecule NPR-A binding site; and

(ii) at least one modifying moiety conjugation site; and

(b) at least one modifying moiety attached to said modifying moiety conjugation site;

wherein said natriuretic compound conjugate is more hydrophilic than a corresponding unconjugated natriuretic compound.

159. A natriuretic compound conjugate comprising:

(a) a natriuretic compound comprising:

(i) a natriuretic molecule NPR-A binding site; and

(ii) at least one modifying moiety conjugation site; and

(b) at least one modifying moiety attached to said modifying moiety conjugation site;

wherein said natriuretic compound conjugate is more amphiphilic than a corresponding unconjugated natriuretic compound.

160. A natriuretic compound conjugate comprising:

(a) a natriuretic compound comprising:

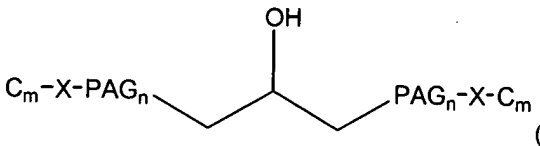
(i) a natriuretic molecule NPR-A binding site; and

(ii) at least one modifying moiety conjugation site; and

(b) at least one modifying moiety attached to said modifying moiety conjugation site;

wherein the natriuretic compound conjugate is more lipophilic than a corresponding unconjugated natriuretic compound, wherein at least one modifying moiety does not consist of an alkyl moiety.

161. A compound having a formula:



(Formula IV)

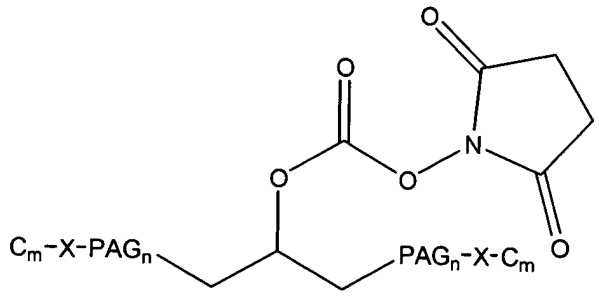
wherein

each C is independently selected and is an alkyl moiety having m carbons and m is from 1 to 20; and

each PAG is independently selected and is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

each X is independently selected and is a linking moiety.

162. A compound having a formula:



(Formula V)

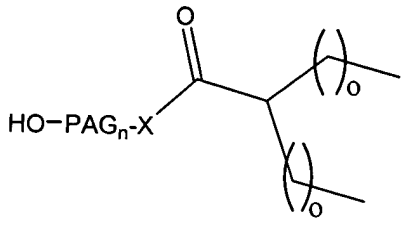
wherein

each C is independently selected and is an alkyl moiety having m carbons and m is from 1 to 20; and

each PAG is independently selected and is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

each X is independently selected and is a linking moiety.

163. A compound having a formula:



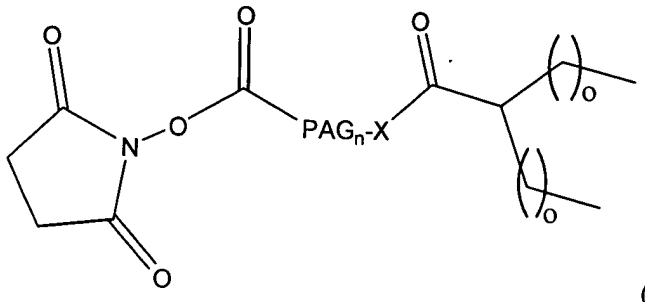
(Formula VI)

PAG is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

X is O or N; and

each o is independently selected and is from 1 to 15.

164. A compound having a formula:



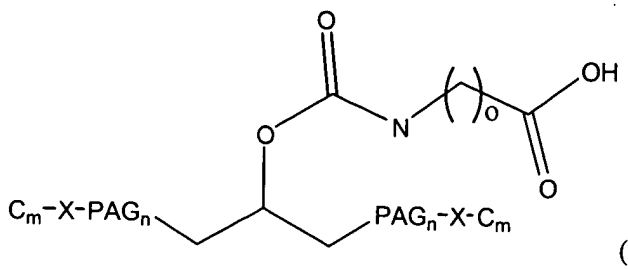
(Formula VII)

PAG is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

X is O or N; and

each o is independently selected and is from 1 to 15.

165. A compound having a formula:



(Formula VIII)

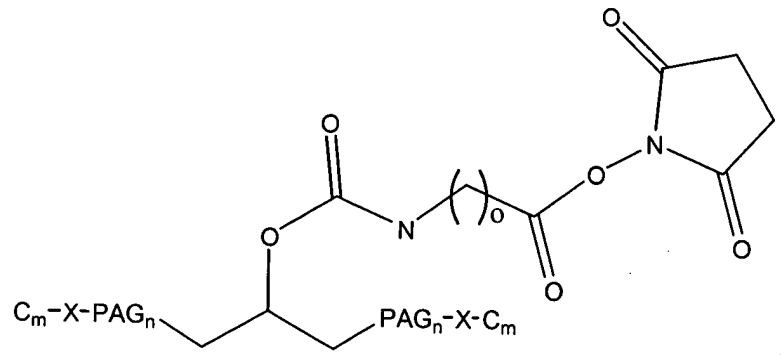
each C is independently selected and is an alkyl moiety having m carbons and m is from 1 to 20; and

each PAG is independently selected and is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

each X is independently selected and is a linking moiety.

o is from 1 to 15.

166. A compound having a formula:



(Formula IX)

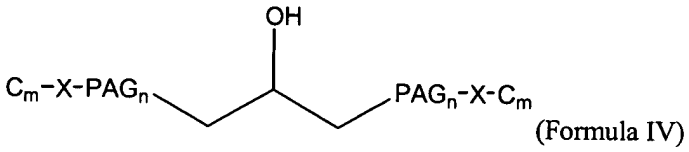
each C is independently selected and is an alkyl moiety having m carbons and m is from 1 to 20; and

each PAG is independently selected and is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

each X is independently selected and is a linking moiety;

o is from 1 to 15.

167. A method of making a compound of the formula:



(Formula IV)

wherein

each C is independently selected and is an alkyl moiety having m carbons and m is from 1 to 20; and

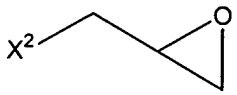
each PAG is independently selected and is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

each X is independently selected and is a linking moiety;

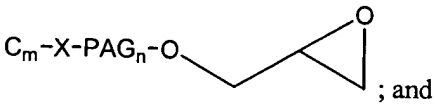
the method comprising:

(a) reacting a compound of formula:

with a compound of formula:



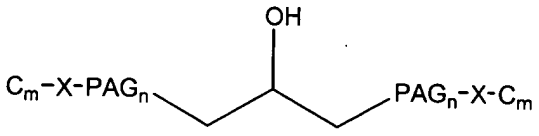
where X^2 is a halide, and wherein the reaction is carried out in the presence of a base and a solvent to yield:



; and

(b) reacting the product of (a) with a compound of formula:

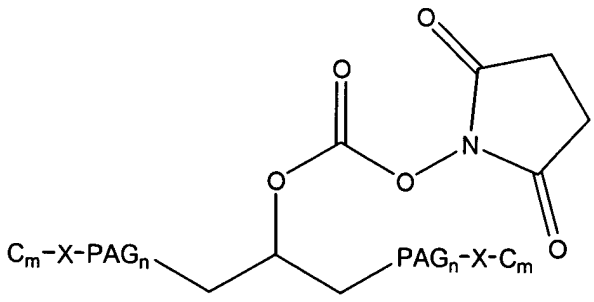
in the presense of a Lewis acid and a solvent to yield:



168. The method of claim 167 wherein the base is NaH and the solvent is tetrahydrofuran.

169. The method of claim 167 wherein the Lewis acid is BF_3OEt_2.

170. A method of making a compound of the formula:



(Formula V)

wherein

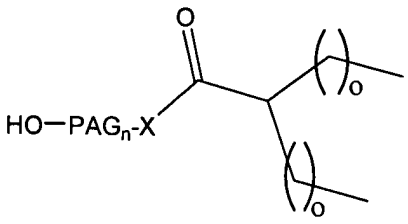
each C is independently selected and is an alkyl moiety having m carbons and m is from 1 to 20; and

each PAG is independently selected and is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

each X is independently selected and is a linking moiety;

the method comprising reacting the product of claim 0 with paranitrochloroformate or disuccimidyl carbonate.

171. A method of making a compound of the formula:



wherein

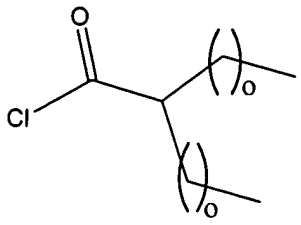
PAG is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

X is O or N; and

each o is independently selected and is from 1 to 15;

the method comprising:

reacting a compound of formula:

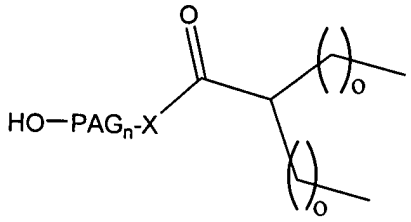


wherein o is as defined above, with a compound of formula:

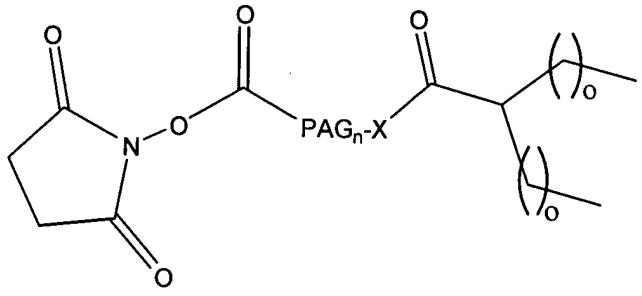
$$HO{-}PAG_n{-}X$$

where X is –NH or –OH;

in solvent, to yield a compound of formula:



172. A method of making a compound of the formula:



(Formula VII)

wherein

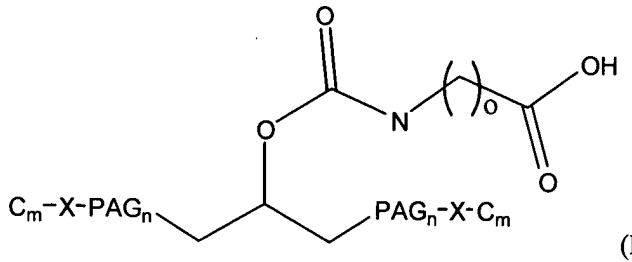
PAG is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

X is O or N; and

each o is independently selected and is from 1 to 15;

the method comprising activating a product of claim 170 using an activating agent selected from the group consisting of disuccinimidyl carbonate, paranitrochloroformate, phosgene and N-hydroxysuccinimide.

173. A method of making a compound of the formula:



(Formula VIII)

wherein

each C is independently selected and is an alkyl moiety having m carbons and m is from 1 to 20; and

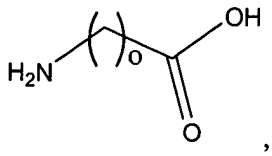
each PAG is independently selected and is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

each X is independently selected and is a linking moiety.

o is from 1 to 15;

the method comprising:

reacting the product of claim 0 with a compound of formula:

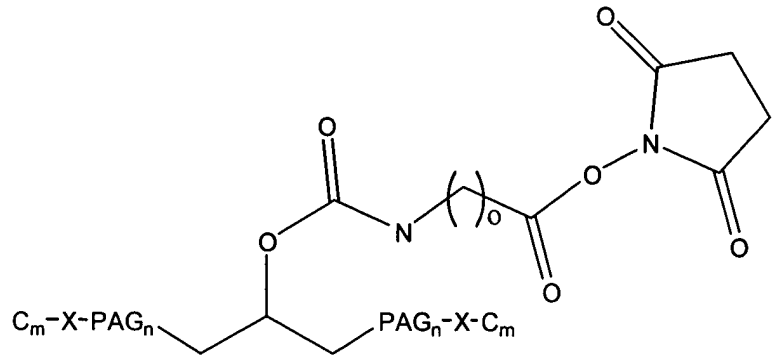


,

in the presence of a base in a solvent.

174. The method of claim 173 wherein the base is K_2CO_3 and the solvent is an aqueous and/or organic solvent.

175. A method of making a compound of the formula:



(Formula IX)

wherein

each C is independently selected and is an alkyl moiety having m carbons and m is from 1 to 20; and

each PAG is independently selected and is a polyalkylene glycol moiety having n subunits and n is from 2 to 25;

each X is independently selected and is a linking moiety;

o is from 1 to 15;

the method comprising reacting a compound produced according to the method of claim 1 with N-hydroxysuccinimide.

176. A natriuretic peptide analog comprising a structure:

SP**X**^{1}MMH**X**^{2}SGCFGRRLDRIGSLSGLGCNVLR**X**^{3}Y

wherein X^1 is Lys, Arg or His, X^2 is Lys, Arg, His, and X^3 is Arg or His.

177. The natriuretic peptide analog of claim 176 comprising a modifying moiety conjugated at the S residue.

178. A natriuretic peptide analog comprising a structure:

SP**Z**^{1}MVQGSG-CFGR**Z**^{2}MDRISSSS**X**^{1}**X**^{2}**X**^{3}C

wherein Z^1 is Arg or an amino acid other than Lys, and wherein Z^2 is Arg or an amino acid other than Lys, wherein X^1 is Gly, Met, Leu, Phe, Ile or a conservative substitution thereof, wherein X^2 is Leu, Trp, Tyr, Phe or a

conservative substitution thereof, and wherein X^3 is Gly and Arg, or a conservative substitution thereof.

179. The natriuretic peptide analog of claim 178 where Z^1 is Lys and Z^2 is other than Lys.

180. A natriuretic peptide analog comprising a structure:

K CFKGKNDR**X**^{1} K**X**^{2} QSGL**X**^{3} C-NSFKY

wherein X^1 is T, a, R, H, P, T, E;

wherein X^2 is K, N-methyl, Arg, S, D,P;

wherein X^3 is Arg, K, Y, F, S, P, Orn, Har, Har, p-amidinophenyl Ala, I, any other amino acid that has a positive charge other than Gly, or Try.

181. The natriuretic peptide of claim 178 or 180 further defined as comprising a natriuretic peptide conjugate, comprising a modifying moiety conjugated to one or more of the Lys residues therein.